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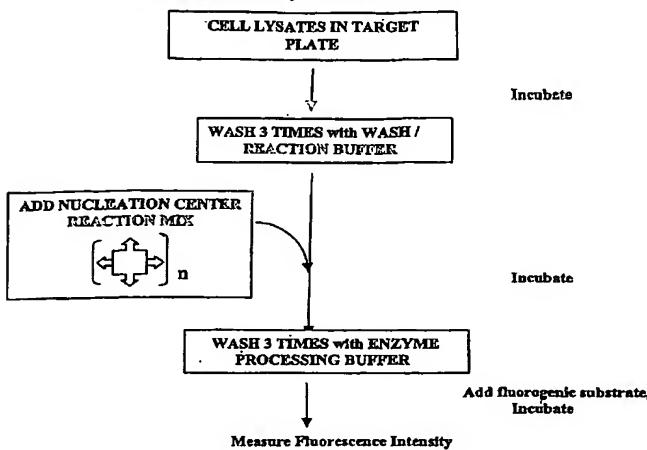
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(54) Title: HOMOGENEOUS AND HETEROGENEOUS ASSAY METHODS USING NUCLEATION CENTERS AND NOVEL COVALENT CHEMISTRIES FOR THE RAPID MEASUREMENT OF PHOSPHORYLATION AND DE-PHOSPHORYLATION

Multi-Step Heterogeneous Assay of the Present Invention



(57) Abstract: Novel chemistries, methods and assays are described for the rapid detection and measurement of phosphorylation or de-phosphorylation of polypeptides, oligonucleotides or lipids based upon the use of phosphoryl, phosphoroamino or phosphorothioate specific hetero-bifunctional linkers combined with fluorescence polarization or fluorogenic detection. Homogeneous assay applications possible using these novel chemistries and methods include in vitro fluorescence polarization assays for the detection of general and isoform specific polypeptide, oligonucleotide and lipid kinase and phosphatase enzymes. Heterogeneous assay applications possible using the same novel chemistries and assay methods enable development of in vitro and in vivo fluorogenic assays of general or specific kinase or phosphatase enzyme activities in cell lysates or serum. In all cases, these new chemistries, methods and assays enable simultaneous multi-kinase detection and profiling.

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